PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY
(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 13664-40PCT	**FOR FURTHER ACTION**	See Form PCT/IPEA/416
International application No. PCT/CA2004/001409	International filing date *(day/month/year)* 26 July 2004 (26-07-2004)	Priority date *(day/month/year)* 25 July 2003 (25-07-2003)
International Patent Classification (IPC) or national classification and IPC IPC(7): C07K 14/62, C07K 1/113, C07K 14/765, A61K 38/28, A61P 3/10, A61P 5/50, A61K 47/42		
Applicant CONJUCHEM, INC. ET AL		

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of 6 sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
   a. [X] *(sent to the applicant and to the International Bureau)* a total of 10 sheets, as follows:
      [X] sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
      [ ] sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. 1 and the Supplemental Box.
   b. [ ] *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s)) ,containing a sequence listing and/or tables related thereto, in electronic form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).
4. This report contains indications relating to the following items:
   - [X] Box No. I Basis of the report
   - [ ] Box No. II Priority
   - [X] Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - [ ] Box No. IV Lack of unity of invention
   - [X] Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - [ ] Box No. VI Certain documents cited
   - [ ] Box No. VII Certain defects in the international application
   - [ ] Box No. VIII Certain observations on the international application

Date of submission of the demand 18 May 2005 (18-05-2005)	Date of completion of this report 29 November 2005 (29-11-2005)
Name and mailing address of the IPEA/CA Canadian Intellectual Property Office Place du Portage I, C114 - 1st Floor, Box PCT 50 Victoria Street Gatineau, Quebec K1A 0C9 Facsimile No.: 001(819)953-2476	Authorized officer Colleen MacFarlane (819) 997-4614

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/CA2004/001409

Box No. I Basis of the report

1. With regard to the language, this report is based on:

[X] the international application in the language in which it was filed

[ ] a translation of the international application into , which is the language of a translation furnished for the purposes of:

[ ] international search (Rules 12.3(a) and 23.1(b))

[ ] publication of the international application (Rule 12.4(a))

[ ] international preliminary examination (Rules 55.2(a) and/or 55.3(a))

2. With regard to the elements of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report):*

[ ] the international application as originally filed/furnished

[X] the description:

[X] pages 2-26 as originally filed/furnished

[X] pages* 1 and 1A received by this Authority on 18 May 2005

[ ] pages* received by this Authority on

[X] the claims:

[ ] pages as originally filed/furnished

[ ] pages* as amended (together with any statement) under Article 19

[X] pages* 27-31 received by this Authority on 18 May 2005

[X] pages* 32-34 received by this Authority on 26 October 2005

[ ] the drawings:

[ ] pages as originally filed/furnished

[ ] pages* received by this Authority on

[ ] pages* received by this Authority on

[X] a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing.

3. [X] The amendments have resulted in the cancellation of:

[X] the description, pages 1

[X] the claims, Nos. 1-38

[ ] the drawings, sheets/figs

[ ] the sequence listing *(specify):*

[ ] any table(s) related to sequence listing *(specify):*

4. [ ] This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).

[ ] the description, pages

[ ] the claims, Nos.

[ ] the drawings, sheets/figs

[ ] the sequence listing *(specify):*

[ ] any table(s) related to sequence listing *(specify):*

* *If item 4 applies, some or all of those sheets may be marked "superseded."*

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/CA2004/001409

Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

The question whether the claimed invention appears to be novel, to involve an inventive step (to be non obvious), or to be industrially applicable have not been examined in respect of:

[ ] the entire international application

[X] claims Nos. 23-31

because:

[X] the said international application, or the said claims Nos. 23-31
relate to the following subject matter which does not require an international preliminary examination *(specify)*:

Although claims 23-31 encompass a method of treatment of the human/animal body which this Authority is not required to examine under Rule 67.1(iv) of the PCT, the IPRP has been established on the basis of the alleged effects of the polypeptides referred to therein.

[ ] the description, claims or drawings *(indicate particular elements below)* or said claims Nos.
are so unclear that no meaningful opinion could be formed *(specify)*:

[ ] the claims, or said claims Nos.
by the description that no meaningful opinion could be formed *(specify)*: are so inadequately supported

[ ] no international search report has been established for said claims Nos.

[ ] a meaningful opinion could not be formed without the sequence listing; the applicant did not, within the prescribed time limit:

[ ] furnish a sequence listing on paper complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Preliminary Examining Authority in a form and manner acceptable to it.

[ ] furnish a sequence listing in electronic form complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Preliminary Examining Authority in a form and manner acceptable to it.

[ ] pay the required late furnishing fee for the furnishing of a sequence listing in response to an invitation under Rules 13*ter*.1(a) or (b) and 13*ter*.2.

[ ] a meaningful opinion could not be formed without the tables related to the sequence listings; the applicant did not, within the prescribed time limit, furnish such tables in electronic form complying with the technical requirements provided for in Annex C-*bis* of the Administrative Instructions, and such tables were not available to the International Preliminary Examining Authority in a form and manner acceptable to it.

[ ] the tables related to the nucleotide and/or amino acid sequence listing, if in electronic form only, do not comply with the technical requirements provided for in Annex C-*bis* of the Administrative Instructions.

[ ] See Supplemental Box for further details.

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Claims	1-37	YES
	Claims	NONE	NO
Inventive step (IS)	Claims	1-37	YES
	Claims	NONE	NO
Industrial applicability (IA)	Claims	1-37	YES
	Claims	NONE	NO

2. Citations and explanations (Rule 70.7)

Reference is made to the following documents:
D1: WO 95/05187 A1 (UNITED MEDICAL 7 DENTAL SCHOOLS OF GUY'S AND ST. THOMAS HOSPITALS)
D2: CA 2334859 A1 (KINGS COLLEGE LONDON; DEUTSHES WOLLFORSCHUNGS INSTITUT)
D3: JONASSEN et al. "Fatty acid acylated insulins display protracted action due to binding to serum albumin." PEPTIDE SCIENCE: PRESENT AND FUTURE PROCEDINGS OF THE INTERNATIONAL PEPTIDE SYMPOSIUM, 1ST KYOTO, NOV. 30 1997 (1999), MEETING DATE 197, pages 674-677. EDITOR: SHIMONISHI, YASUTSUGA. PUBLISHER: KLUWER, ORDRECHT, NETH.
D4: BAUDYS et al. "Extending insulin action in vivo by conjugation to carboymethyl dextran." BIOCONJUGATE CHEM. 1998, vol 9, pages 176-183.
D5: CA 2363712 A1 (CONJUCHEM INC.)

NOVELTY

The instant invention is an insulin derivative comprising an insulin molecule and a reactive group (α,β-unsaturated carbonyl moiety, a succinimidyl-containing group or a maleimido-containing group) for covalently bonding a blood component so as to prolong insulin activity and reduce the number of injections necessary to maintain blood glucose levels in glycemic-related conditions.

Document D1 discloses insulin analogues comprising insulin, or a functional equivalent thereof, conjugated to a pendant molecule at the B1 residue which has an affinity for binding proteins in blood plasma, with thyroxine exemplified as a pendant molecule, to treat glycemic-related diseases. Similarly, D2 discloses an insulin analogue comprising insulin conjugated to 3,3',5-triiodothyroxine at the B1 position allowing for binding with thyroxine binding proteins. D3 discloses the acylation of insulin at the B29 position by fatty acids allowing binding to serum albumin and its use in the treatment of diabetes. D4 discloses an insulin conjugate comprising carboxymethyl dextran (CMD) attached to Gly A1 of insulin which allows the binding of 3-4 insulin molecules (a blood component) to one CMD chain so as to stabilize and prolong insulin action. Finally, D5 discloses a method of derivatizing insulinotropic peptides (GLP-1 and exendin 3 and 4) with reactive groups (maleimido and succinimidyl) with or without a linker so as to bind blood components for the purpose of prolonging the insulinotropic activity. While D1-D4 each discloses insulin derivatives capable of binding blood components for the purpose of prolonging insulin activity *in vivo* to effectively treat glycemic-related disorders and D5 teaches the derivatization of insulin-related hormones with maleimido and succinimidyl, none of the documents disclose an insulin derivative conjugated to the aforementioned reactive groups allowing the covalent bonding of the derivative to blood components. Claims 1-37 are therefore considered novel under Article 33(2) of the PCT.

Continued in Supplemental Box

Supplemental Box relating to Sequence Listing

Continuation of Box No.1, item 2:

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, this report was established on the basis of:

   a. type of material

      [X] a sequence listing

      [ ] table(s) related to the sequence listing

   b. format of material

      [X] on paper

      [X] in electronic form

   c. time of filing/furnishing

      [ ] contained in the international application as filed

      [ ] filed together with the international application in electronic form

      [X] furnished subsequently to this Authority for the purposes of search and/or examination

      [ ] received by this Authority as an amendment* on

2. [ ] In addition, in the case that move than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

* *If item 4 in Box No. 1 applies, the listing and/or table(s) related thereto, which form part of the basis of the report, may be marked "superseded".*

Supplemental Box

In case the space in any of the preceding boxes is not sufficient.

Continuation of: Box V

INVENTIVE STEP

Although the prior art discloses insulin derivatives capable of binding blood components and the therapeutic advantages thereof (D1-D4) as well as the derivatization of insulinotropic hormones with maleimido and succinimidyl groups so as to covalently bond blood components for the purpose of protracted activity (D5), a skilled person would not necessarily be lead directly and without difficulty to the instant insulin derivatives capable of covalently bonding blood components. Claims 1-37 are therefore considered inventive under Article 33(3) of the PCT.

INDUSTRIAL APPLICABILITY

Claims 1-22 and 32-36 appear to define subject matter that has industrial applicability under Article 33(4) of the PCT, based on the function of the insulin derivatives of the instant application.

For the assessment of claims 23-31 on the question of whether or not they define subject matter that has industrial applicability, no unified criteria exists in the PCT. Further, the patentability of said claims can depend upon their formulation. Although methods per se defined in claims 23-31 relate to subject matter which this Authority is not obliged to examine under Rule 67.1(iv) of the PCT, the use of the compounds referred to therein for the treatment of glycemic-related disorders appears to represent subject matter that has industrial applicability.

LONG LASTING INSULIN DERIVATIVES AND METHODS THEREOF

BACKGROUND OF THE INVENTION

(a) Field of the Invention

[0001] This invention relates to a long lasting insulin derivative. More particularly, the insulin derivative comprises an insulin molecule and a reactive group coupled thereto, the reactive group being for covalently bonding a blood component hence generating a long lasting insulin derivative.

(b) Description of Prior Art

[0002] Insulin is a vital endocrine hormone that binds to a cellular surface receptor setting off a cascade of events culminating in glucose absorption from the blood. Impaired levels of insulin lead to severe disorders such as types I and II diabetes. Type I diabetes is a life threatening disease where the patient must daily self-administer multiple doses insulin for survival. Type II diabetes, is also a severe medical disease where the endogenous levels of insulin can no longer maintained correct levels of glycemia because the patient due to a tolerance developed by the patient to endogenous levels of insulin. In order to reduce the onset of long-term consequences, a treatment with insulin becomes necessary after failure in lifestyle changes or when traditional glycemia controlling drugs become ineffective. International Patent Application WO 95/05187 describes insulin or a functional derivative equivalent thereof which is covalently linked to a pendant molecular group, which has an affinity for a binding protein. The binding of the pendant molecular group and the binding protein is not covalent. Such binding forces may be for instance electrostatic (eg attraction of opposite charges, hydrogen bonding) or hydrophobic. Therefore, such compounds may not be appropriate for providing a long-lasting effect. Canadian Patent Application 2,363,712 relates to stimulators of insulin release. In particular, this application describes a long-lasting insulinotropic compounds, which stimulates release of a patient's endogenous insulin in response to stimuli. The need that such an insulinotropic compound satisfies is thus to allow or enhance release of endogenous insulin by pancreatic cells in response to

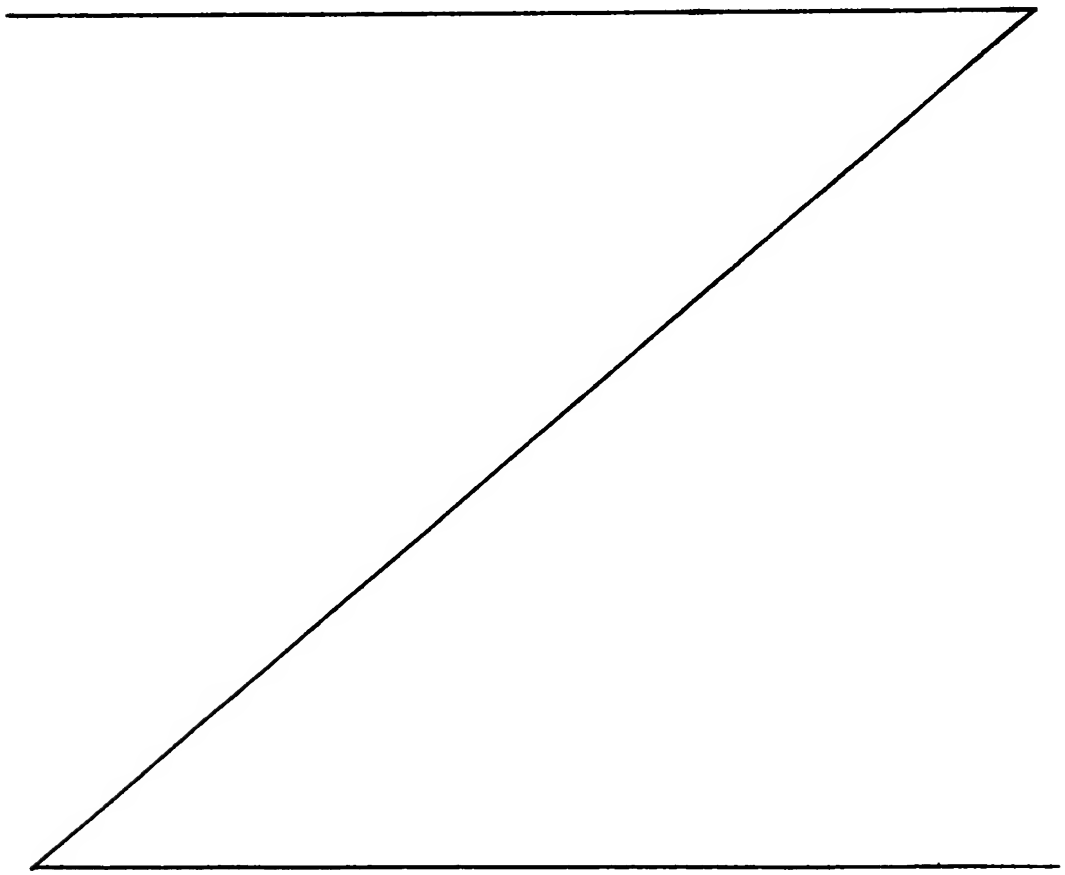
a stimuli during a long period of time. However, this document fails to teach a long-lasting insulin derivatives or a long-lasting exogenous insulin.

[0003] Success in the control of glycaemic disorder is highly related with the compliance of patients to the treatment, and reducing the frequency of injection needed is desirable. To do so, it would be highly desirable to be provided with a new long lasting insulin derivative.

SUMMARY OF THE INVENTION

[0004] In accordance with the present invention there is provided an insulin derivative comprising an insulin molecule and a reactive group for covalently bonding a blood component.

[0005] In a preferred embodiment of the present invention, the insulin molecule is of formula I:

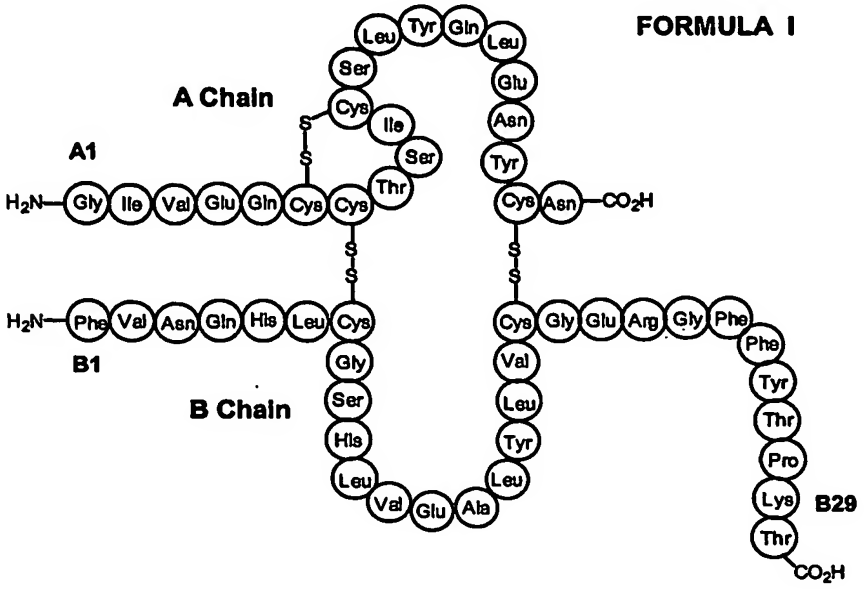


WHAT IS CLAIMED IS:

1. An insulin derivative comprising an insulin molecule and a reactive group for covalently bonding a blood component, said reactive group being selected from the group consisting an α,β-unsaturated carbonyl moiety, a succinimidyl-containing group and a maleimido-containing group.

2. The insulin derivative of claim 1, wherein the insulin molecule is of formula I:

FORMULA I



and the reactive group is coupled to an amino acid of the insulin molecule at a position selected from the positions Gly A1, Phe B1 and Lys B29.

3. The insulin derivative of claim 1 or 2, wherein the reactive group is a maleimido-containing group.

4. The insulin derivative of claim 1 or 2, wherein the reactive group is 3-Maleimidopropionic acid (MPA).

5. The insulin derivative of any one of claims 1 to 4, wherein the reactive group is coupled to an amino acid of the insulin molecule via a linker.

6. The insulin derivative of claim 5, wherein said linker is selected from the group consisting of (2-amino) ethoxy acetic acid (AEA), ethylenediamine (EDA), amino ethoxy ethoxy succinimic acid (AEES), AEES-AEES, 2-[2-(2-amino)ethoxy)] ethoxy acetic acid (AEEA), AEEA-AEEA, $-NH_2-(CH_2)_n-COOH$ where n is an integer between 1 and 20 and alkyl chain (C1-C10) motif and combination thereof.

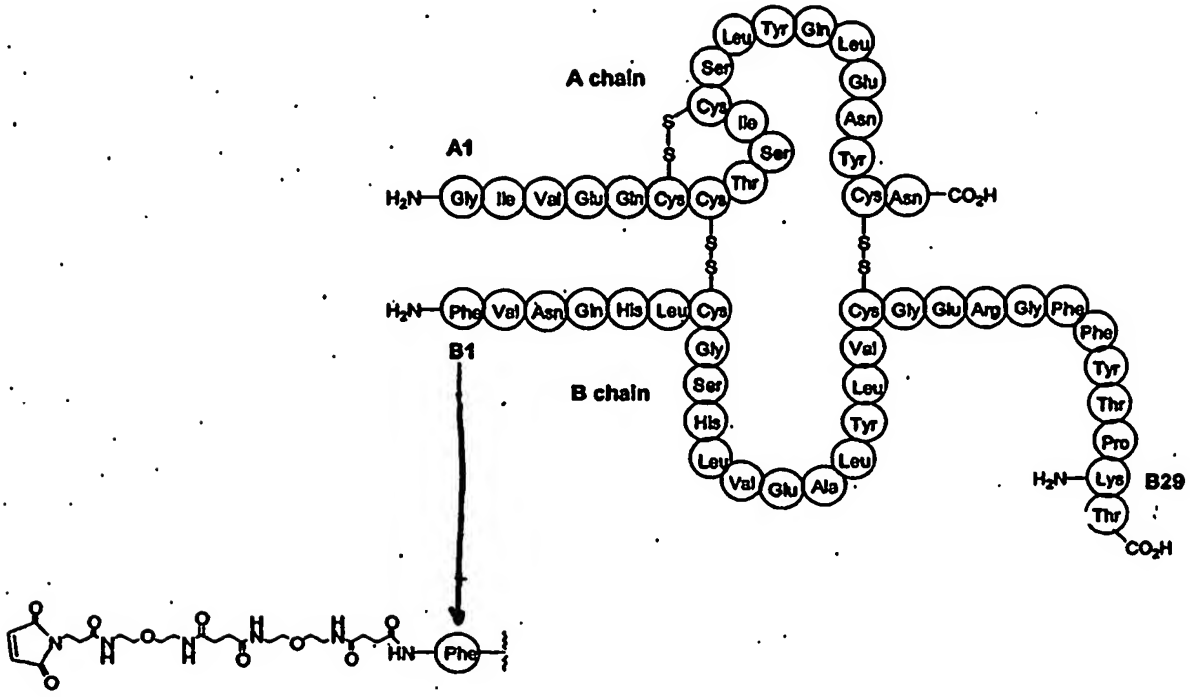
7. The insulin derivative of claim 6, wherein said alkyl chain (C_1-C_{10}) motif is one or more alkyl chains (C_1-C_{10}) saturated or unsaturated in which could be incorporated oxygen nitrogen or sulfur atoms.

8. The insulin derivative of claim 7, wherein said alkyl chain is selected from the group consisting of glycine, 3-aminopropionic acid (APA), 8-aminooctanoic acid (AOA) and 4-aminobenzoic acid (APhA).

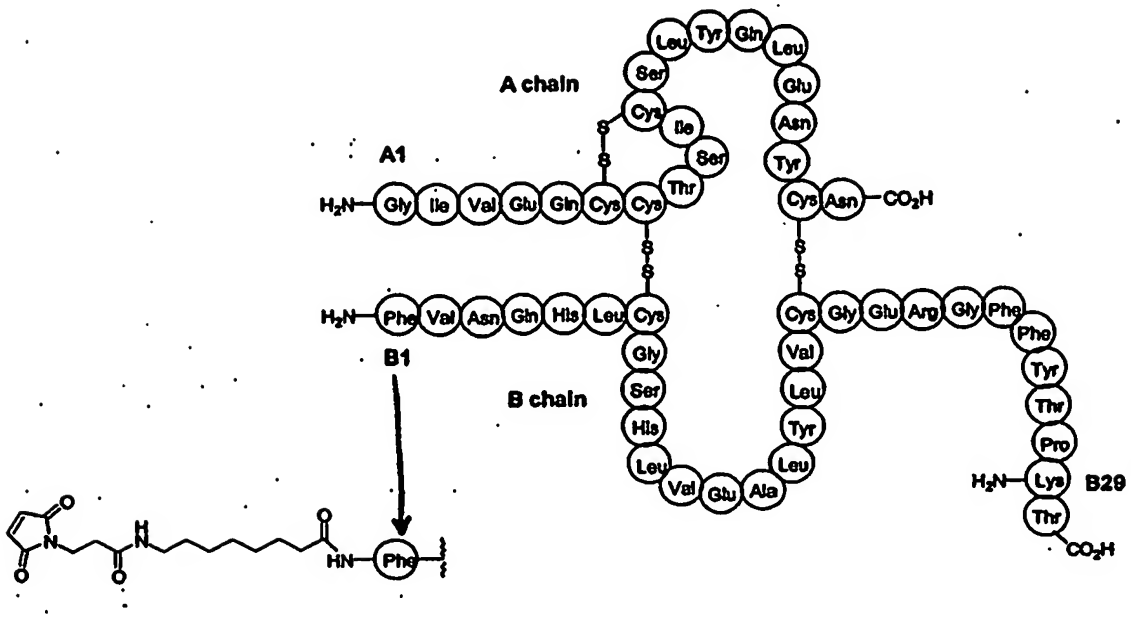
9. The insulin derivative of claim 6, wherein said combination is selected from the group consisting of AEEA-EDA, AEEA-AEEA and AEA-AEEA.

10. The insulin derivative of claim 6, wherein said linker is $-NH_2-(CH_2)_7-COOH$.

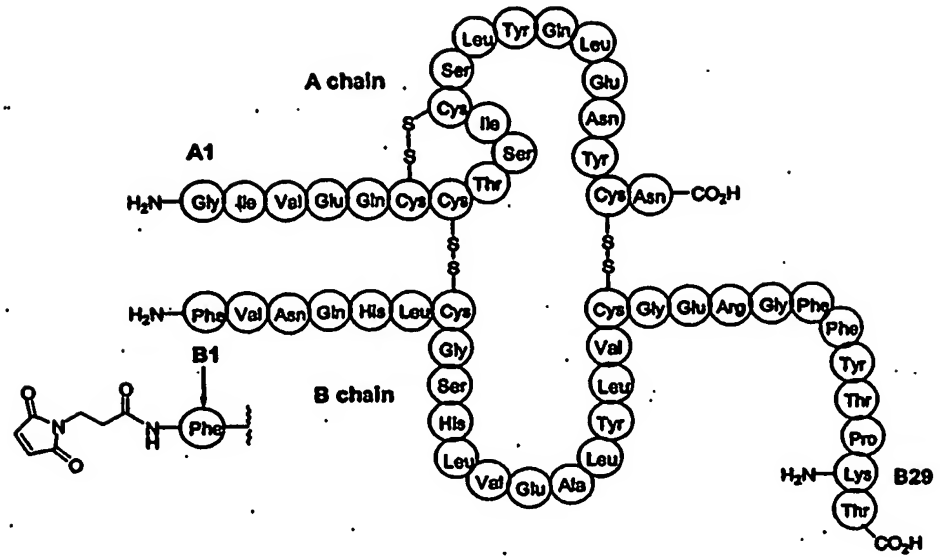
11. The insulin derivative of claim 1 having the formula:



12. The insulin derivative of claim 1, having the formula:



13. The insulin derivative of claim 1, having the formula:



14. The insulin derivative of claim 1, wherein said blood component is a blood protein.

15. The insulin derivative of claim 14, wherein said blood protein is serum albumin.

16. An insulin conjugate comprising an insulin derivative according to any one of claims 1 to 15 and a blood component, wherein the reactive group and the blood component are conjugated through a covalent bond formed between said reactive group and said blood component.

17. The insulin conjugate of claim 16, wherein the blood component is a blood protein.

18. The insulin conjugate of claim 17, wherein the blood protein is serum albumin.

19. The insulin conjugate of claim 16, wherein said conjugate was formed *ex vivo*.

20. The insulin conjugate of claim 19, wherein said blood component is recombinant albumin.

21. A pharmaceutical composition comprising the insulin derivative of any one of claims 1 to 15 in association with a pharmaceutically acceptable carrier.

22. A pharmaceutical composition comprising the insulin conjugate of any one of claims 16 to 20 in association with a pharmaceutically acceptable carrier.

23. A method for treating a glycaemic-related disease or disorder in a subject suffering from said glycaemic-related disease or disorder, comprising administering the insulin derivative of any one of claims 1 to 15 to said subject.

24. The method according to claim 23, wherein said glycaemic-related disease is selected from the group consisting of diabetes of type I, diabetes of type II, gestational diabetes, cystic fibrosis, polycystic ovary syndrome and pancreatitis.

25. The method according to claim 23, wherein the glycaemic-related disease is selected from the group consisting of diabetes of type I and diabetes of type II.

26. A method for treating a glycaemic-related disease or disorder, comprising the administration of the insulin conjugate of any one of claims 16 to 20.

27. The method according to claim 26, wherein said glycaemic-related disease is selected from the group consisting of diabetes of type I, diabetes of type II, gestational diabetes, cystic fibrosis, polycystic ovary syndrome and pancreatitis.

28. The method according to claim 26, wherein the glycaemic-related disease is selected from the group consisting of diabetes of type I and diabetes of type II.

29. A method for treating a glycaemic-related disease or disorder, comprising the administration of the pharmaceutical composition of any one of claims 21 and 22.

30. The method according to claim 29, wherein said glycaemic-related disease is selected from the group consisting of diabetes of type I, diabetes of type II, gestational diabetes, cystic fibrosis, polycystic ovary syndrome and pancreatitis.

31. The method according to claim 29, wherein the glycaemic-related disease is selected from the group consisting of diabetes of type I and diabetes of type II.

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32. Use of the derivative of any one of claims 1 to 15, for the preparation of a medicament for the treatment of a glycaemic-related disease or disorder.

33. The use as claimed in claim 32, wherein said glycaemic-related disease is selected from the group consisting of diabetes of type I, diabetes of type II, gestational diabetes, cystic fibrosis, polycystic ovary syndrome and pancreatitis.

34. The use as claimed in claim 32, wherein the glycaemic-related disease is selected from the group consisting of diabetes of type I and diabetes of type II.

35. Use of the conjugate of any one of claims 16 to 20, for the preparation of a medicament for the treatment of a glycaemic-related disease or disorder.

36. The use as claimed in claim 35, wherein said glycaemic-related disease is selected from the group consisting of diabetes of type I, diabetes of type II, gestational diabetes, cystic fibrosis, polycystic ovary syndrome and pancreatitis.

37. The use as claimed in claim 36, wherein the glycaemic-related disease is selected from the group consisting of diabetes of type I and diabetes of type II.